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A

Synopsis

On

"Studies on Agronomical importance of *Trichoderma* Species from saline soil"

For the Degree of Doctor of Philosophy In

Microbiology

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Abstract-

Soil salinity is a pending threat to global agricultural sustainability and food security. Plantbeneficial Microbes positively influence the microbial community in the rhizoshere, and thereforeprotect the plants as biological control agents (BCAs) and stimulate their growth as plant growthpromotingmicrobes (PGPMs).*Trichoderma* is considered to be the mostbeneficial fungal genusfor solving Agricultural problems with very less adverse effects. The agronomical importance of*Trichoderma* lies in its potential as biocontrol against phytopathogens , Production of range of enzymes, bioremediation.Present study will focus on Isolation, Screening, & identification of salinity tolerant *Trichoderma* from saline agricultural soils and Mangrove soils of Maharashtra ,India. The study will also explore the possibilities of its applications in direct and indirect plant growth promotion, bioremediation of agro-waste.

1] INTRODUCTION:

Soil salinity refers to the presence of high concentrations of soluble saltsin the soil moistureof the root zone. These concentrations of soluble salts, through their high osmotic pressures, affectplant growth by restricting the uptake of water by the roots. All plants are subject to this influence,but sensitivity to high osmotic pressures varies widely among plant species. Salinity can alsoaffect plant growth because the high concentration of salts in the soil solution interferes with abalanced absorption of essential nutritional ions by the plants. The proliferation of plant pathogenicfungi that cause damping off, wilt, and root-rot diseases are the principal effects of salinity on plantgrowth and agricultural production. Physiologic drought, withering, and smaller seeds; insufficientseed germination; Weed plants that are salt-tolerant or halophilic; Low yields of seeds and other plant parts are a result of all of the senegative circumstances.(Parida and Das2005).

Biological control offers the chance toimprove crop production within the existing resources,besides avoiding the problem of pesticide resistance (Khan et al., 2014).However; it has been considered as a more natural and environmentally acceptable alternative to existing chemical treatment methods (Baker and Paulitz,1996;Eziashi etal.,2007)

The utilization of adaptable microorganisms becomes a novel strategy to abiotic stress relief.*Trichoderma* spp. has a variety of functions and capacities in terms of favorable plant connections (Hidangmayum and Dwivedi, 2018). Rice seed priming with *Trichoderma* promotes root and shoot growth,water and nutrient uptake, and plant resilience (Rawatetal.,2012).It also improves salt tolerance, lowering agronomic parameter stress (Rawat et al., 2016; Yasmeen and Siddiqui, 2017). Application of *Trichoderma* reduces the adverse effects of salinity stress on wheat growth (Zhang et al., 2016; Zhang et al., 2019), and French bean (Gupta and Pandey,2019). *Trichoderma* are closely related with, to degrade substrates and to possess high resistance to microbial inhibitors (Strange1993)

Trichoderma spp. include a number of fungal strains that are used as biocontrol agents due to their abilities to antagonize a wide range of phyotopathogenic fungi ,bacteria.

2] Review of Literature and Development in the subject (Previous work done in the relevant area):

□ **International status-**

Salt tolerant *T.atroviride* isolated from marine habitat having antagonistic activity to F.*oxysporum*. And it can be used as biological control agent in saline alkali land. (Chong yuan zhan, Weiweiwang etal.,2022) China.

Trichoderma species of salt tolerating potential play the role of natural biological control.Emerging salt marshes as a source of *Trichoderma arenarium* sp.nov.and other fungal bioeffectors for biosaline agriculture.(Ming.Yue Ding,Wei etal.,2021) china

Effect of salinity on growth of *Trichoderma herzianum* having antigonastic activity. (Boumaaza Boualem etal.,2015) Algeria.

The potent fungus *Trichoderma* species which able to produce natural compounds vital for disease

Resistance of soil ,which spread dramatically In the saline soil and cause significant loss in agricultural crops.(Eman R Hamed Awad etal.,2015) Egypt.

The mycosynthesis of several metalnanoparticles from *Trichoderma* species for use in agri-food applications.The fungal-cell or cell-extract –derived NPs (mycogenic NPs) can be applied as nanofertilizers, nanofungicides, plant growth stimulators, nanocoatings (Mousa A. Alghuthaymi, Kamel A. Abd-Elsalam etal.,**2022**,*8*,367.https://doi.org/10.3390/jof8040367) Egypt.

□**National Status**

High Salinity tolerant *Trichoderma* species having plant growth property and antagonistic activity against various soil born phytopathogens and identification of the fungus using biochemical characteristics. (Viveksingh,Chetankeswani.,2019) Varanasi,India

Bioremediation of salt affected soils by the potent fungal spp. (sanjay arora, Atul K. sing., 2017) Lakhnow,India

Production and characterization of IAAby the isolate *T. viride* VKF3 from Mangrove associated area. The plant growth promotional activity as well as its biocontrol efficacy against the fungal pathogens.(NathanVinod Kumar,K.Subha Rajamand Mary Esther Rani.,2017)Tamilnadu,India

Trichoderma can be used for the Production of laccase have application in dye decolourisation (Ranimol.G,Thulasi Venugopal,Shiji Gopala krishnan,Swetha Sunkar.,2018)Tamilnadu, India

Some important antagonistic fungi Produces siderophores, involved in plant growth promotion.(Swapan Kr.Ghosh,Subhankar Banerjee and Chandan Sengupta.,2017)

Different species of *Trichoderma* isolates were used for production of nanoparticles and characterization of the synthesized silver nanoparticles for solving enverionmental problems (T Prameela Devi Kulanthaivel etal.,2013) New Delhi 110 012, India

3. Objectives of Research /Proposed Hypothesis:

1) To collect soil samples from saline soils and mangrove soils of Maharashtra.
2) To isolate *Trichoderma spp.*from different saline soil samples.
3) To study the antagonism of selected *Trichoderma* against soil born fungal phytopathogens.
4) To Screen the different *Trichoderma* isolates for salt tolerance potential.
5) To identify the *Trichoderma* isolates by PCR& Sequencing.
6) To study the Plant growth promoting traits of *Trichoderma* isolates such as
    - Phosphate Solubilization Potentials
    - Siderophore production
    - Production of Indole-3-Acetic acid(IAA)
    - Fungal cell wall degrading enzyme activity
7) To screen the production of Cellulase and Laccase
8) To synthesize nanoparticals from *Trichoderma* isolates.
9) Characterization of *Trichoderma* nanoparticles.
10) Antimicrobial activity of *Trichoderma* nanoparticles,
11) Qualitative Study of selected *Trichoderma* isolate(s) potential for biodegradation of cellulosic agro-waste
12) Pottrials for plant growth promoting potential by *Trichoderma* isolates in Wheat (Triticum aestivum) and Gram (Cicer arietinum)

4. Methodology tobeadopted:

1) Isolation of different types of *Trichoderma* spp from saline soil samples collected from costallines of Maharashtra using potato dextrose agar,rose bengol agar and other selective media

2) Screening of isolated *Trichoderma* spp. For salt tolerance by growing at various salt concentrations.

3) Identification & Characterization (Morphological&Molecular) of different Isolates by using standard protocol or techniques.

4) Antagonism effect of identified *Trichoderma* against soil born fungal phytopathogens on PDA medium.

5) Estimation of plant growth promoting traits of identified *Trichoderma* spp.such as

- Phosphate solubilization potential using Pikovaskys agar medium and broth.
- Siderophore production by Universal Chemical assay i.e.Chrome Azurol Sulphonate (CAS) assay.

  The detection of siderophore type will be carried out by UV-Visible spectroscopy, the Csaky test for hydroxamate type and Arnow's test for catecholate-phenolate type of siderophore
- Chitinase activity using colloidal Chitin amended medium.Microscopic observation will be used to detect degradation of pathogenic fungal cell wall.
- For cellulose production carboxy methyl cellulose amended medium will be used for qualitative and quantitative determination
- Laccase producers will be screened on malt extract agar (MEA) plates containing 0.01%(w/v) guaiacol or 2, 20-azino-bis (3-ethyl benzo thiazo line-6-sulfonic acid) [ABTS] as an indicator compound to evaluate the presence of laccase activity.
- Synthesis of silver nanopartical From *Trichoderma* isolates will be carried out using 1m M silver nitrate solution in mycelia free cell filtrate (MFCF)
- Characterization of silver nanoparticles (AgNPs) by UV–visible spectra, Scanning electron microscopy (SEM), X-ray diffraction (XRD),FTIR analysis.
- Antimicrobial activity of *Trichoderma* nanoparticals against soil born fungal Phytopathogens on PDA medium.
- Rate of cellulosic agro-waste decomposition by selected *Trichoderma* strain(s) will be tested comparing with traditional method
- Pot trials for plant growth promoting potential by *Trichoderma* isolates in Wheat(Triticum aestivum) and Gram(Cicer arietinum) will be conducted using seed treatment as well as foliar application of secondary metabolites.

4. Importance of study /Society application:

□ **Significance of the study-**

The farming land is declining gradually and the main reasons are intensive use of agricultural practices, urbanization, biotic and abiotic stress etc. Among abiotic stresses the salinity problem is increasing at an alarming rate throughout the world.Salinity is responsible for the loss of crop production every year.

The salt tolerant ,plant growth promoting *Trichoderma* strain may provide an ecofriendly solution to the problems related to salt stress induced lowered crop yields, provision of nutrients protection against fungal phytopathogens and biodegradation of cellulosic agro-waste.

5. ProposedworkPlan/FormulationandStructureofStudy:

□ **Year-wisePlanofworkandtargetstobeachieved**

Thefollowingplanofstudyhasbeenproposedto accomplishtheaimsand objectivesofthepresentstudy.

* **FirstYearplanofwork**

1) Study the course-work for research awareness and to enhance the self growth with respect to concept and knowledge of the subject.
2) Extensive Review of literature related to the topic
3) The collection and characterization of different saline soil samples.
4) Isolation of *Trichoderma spp.*

* **SecondYearplanofwork-**

1) Screening of salt tolerant *Trichoderma* isolates
2) Screening of *Trichoderma* isolates for plant growth promoting traits
3) Conventional and molecular characterization of *Trichoderma* isolates
4) Standardization of media for production of IAA and Siderophore

* **ThirdYearplanofwork-**

1) Study of Antagonism of selected *Trichoderma* isolate against various soil bornphytopathogens.
2) Synthesis of nanoparticals from *Trichoderma* isolates.
3) Characterization of *Trichoderma* nanoparticles.
4) Antimicrobial activity of *Trichoderma* nanoparticles on phytopathogens.
5) Study of Cellulase and Laccase production
6) Studyof Cellulosic agro-waste decomposition
7) Pot trials for plant growth promoting potential by *Trichoderma* isolates in Wheat (Triticum aestivum)andGram(Cicer arietinum)

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Signature **Signature**

ResearchScholar **ResearchSupervisor/Guide/Co-guide**